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BIOLOGICAL INTEGRITY OF BEAVER CREEK IN THE BIG SPRING CREEK TMDL PLANNING AREA BASED ON THE STRUCTURE AND COMPOSITION OF THE BENTHIC ALGAE COMMUNITY

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Summary

In July 2003, periphyton samples were collected from three sites on Beaver Creek in the Big Spring Creek TMDL planning area in central Montana to assess whether this stream is water-quality limited and in need of TMDLs. The Beaver Creek samples were collected following MDEQ standard operating procedures, processed and analyzed using standard methods for periphyton, and evaluated following modified USEPA rapid bioassessment protocols for wadeable streams.

Samples of conspicuous macroscopic algae were also collected from Casino Creek and Cottonwood Creek in July 2003 to identify the taxa responsible for these growths. Results are reported here and in a memo dated October 9, 2003. These samples were not collected following MDEQ standard operating procedures.

The upper site on **Beaver Creek** was moderately impaired by sedimentation and provided less than full support for aquatic life uses. The pollution index was depressed at the upper site, suggesting minor impairment from organic loading. The lower site was also moderately impaired by sedimentation, but the pollution index here was just above the threshold for minor impairment. The middle site had the best biological integrity of the three sites on Beaver Creek. Diatom metrics indicate only minor disturbance and minor impairment from sedimentation here. The eutraphentic diatom *Gomphonema minutum* was a dominant species at both the middle and lower sites, indicating elevated concentrations of inorganic nutrients at both sites. The green alga *Cladophora* ranked second in biomass at these two sites.

The algal specimen collected from **Casino Creek** was a large colony of *Cladophora*, probably *Cladophora glomerata*. *Cladophora* is a common mat-forming filamentous green alga that often becomes a nuisance in nutrient-rich waters around the world. The bright green color indicates fresh new growth, that is, epiphytic diatoms had not yet had a chance to colonize the filaments.

The algal specimen collected from **Cottonwood Creek** proved to be a colony of diatom cells embedded in amorphous mucilage. The cells were mainly a *Cymbella* species and a *Synedra* species. These taxa are known to extrude polysaccharide mucilage through pores in their cell walls, thus forming stalks, tubes, and amorphous masses in which the diatoms live. Certain species of *Cymbella* and *Synedra* are associated with eutrophication.

Introduction

This report evaluates the biological integrity[1], support of aquatic life uses, and probable causes of stress or impairment to aquatic communities in Beaver Creek, located in the Big Spring Creek TMDL planning area of central Montana. The main purpose of this report is to provide information that will help the State of Montana determine whether Beaver Creek is water-quality limited and in need of TMDLs. This report also describes conspicuous algal growths collected from nearby Casino Creek and Cottonwood Creek.

The federal Clean Water Act directs states to develop water pollution control plans (Total Maximum Daily Loads or TMDLs) that set limits on pollution loading to water-quality limited waters. Water-quality limited waters are lakes and stream segments that do not meet water-quality standards, that is, that do not fully support their beneficial uses. The Clean Water Act and USEPA regulations require each state to (1) identify waters that are water-quality limited, (2) prioritize and target waters for TMDLs, and (3) develop TMDL plans to attain and maintain water-quality standards for all water-quality limited waters.

Evaluation of aquatic life use support in this report is based on the species composition and structure of periphyton (benthic algae, phytobenthos) communities at stream sites that were sampled in July 2003. Periphyton is a diverse assortment of simple photosynthetic organisms called algae that live attached to or in close proximity of the stream bottom. Some algae form long filaments or large colonies and are conspicuous to the unaided eye. But most, including the ubiquitous diatoms, can be seen and identified only with the aid of a microscope. The periphyton community is a basic biological component of all aquatic ecosystems. Periphyton accounts for much of the primary production and biological diversity in Montana streams (Bahls et al. 1992). Plafkin et al. (1989) and Barbour et al. (1999) list several advantages of using periphyton in biological assessments.

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[1] *Biological integrity* is defined as "the ability of an aquatic ecosystem to support and maintain a balanced, integrated, adaptive community of organisms having a species composition, diversity, and functional organization comparable to that of natural habitats within a region" (Karr and Dudley 1981).

Project Area and Sampling Sites

The project area is located in Fergus County in central Montana. Beaver Creek, Casino Creek, and Cottonwood Creek are tributaries of Big Spring Creek, which is a principal tributary of the Judith River. These streams head south of Lewistown on the north side of the Big Snowy Mountains, which is an outlier of the Middle Rockies Ecoregion (USEPA 2000).

Periphyton samples were collected at three sites on Beaver Creek (Table 1). Elevations at the sampling sites range from about 4400 feet above mean sea level at the upper site to about 3840 feet at the lower site. Vegetation in the study area is mainly mixed conifer forest in the upper reaches of Beaver Creek and mixed grassland along the middle and lower reaches (USDA 1976). Land use is primarily livestock grazing. Samples of conspicuous algae were also collected from nearby Casino Creek and Cottonwood Creek (Table 1).

Methods

Periphyton samples were collected from Beaver Creek following standard operating procedures of the MDEQ Planning, Prevention, and Assistance Division. Using appropriate tools, microalgae were scraped, brushed, or sucked from natural substrates in proportion to the importance of those substrates at each study site. Macroalgae were picked by hand in proportion to their abundance at the site. All collections of microalgae and macroalgae were pooled into a common container and preserved with Lugol's (IKI) solution. Grab samples of conspicuous macroalgae (for identification only) were collected from Casino Creek and Cottonwood Creek and preserved with Lugol's solution.

The Beaver Creek samples were examined to estimate the relative abundance and rank by biovolume of diatoms and genera of soft (non-diatom) algae according to the method described in Bahls (1993). Soft algae were identified using Smith (1950), Prescott (1962, 1978), John et al. (2002), and Wehr and Sheath (2003). These books also served as references on the ecology of the soft algae, along with Palmer (1969, 1977).

After the identification of soft algae, the raw periphyton samples were cleaned of organic matter using sulfuric acid, postassium dichromate, and 3% hydrogen peroxide. Then, permanent diatom slides were prepared using Naphrax, a high refractive index mounting medium, following *Standard Methods for the Examination of Water and Wastewater* (APHA 1998). Approximately 350 diatom cells (700 valves) were counted at random and identified to species. The following were the main taxonomic references for the diatoms: Krammer and Lange-Bertalot 1986, 1988, 1991a, 1991b; Lange-Bertalot 2001; Krammer 2002. Diatom naming conventions followed those adopted by the Academy of Natural Sciences for USGS NAWQA samples (Morales and Potapova 2000). Van Dam et al. (1994) was the main ecological reference for the diatoms.

The diatom proportional counts were used to generate an array of diatom association metrics. A metric is a characteristic of the biota that changes in some predictable way with increased human influence (Barbour et al. 1999). Diatoms are particularly useful in generating metrics because there is a wealth of information available in the literature regarding the pollution tolerances and water quality preferences of common diatom species (e.g., Lowe 1974, Beaver 1981, Lange-Bertalot 1996, Van Dam et al. 1994).

Values for selected metrics were compared to biocriteria (numeric thresholds) developed for streams in the Rocky Mountain ecoregions of Montana (Table 2). These criteria are based on the distribution of metric values measured in least-impaired reference streams (Bahls et al. 1992) and metric values measured in streams that are known to be impaired by various sources and causes of pollution (Bahls 1993). The biocriteria in Table 2 are valid only for samples collected during the summer field season (June 21-September 21).

The criteria in Table 2 distinguish among four levels of stress or impairment and three levels of aquatic life use support: (1) no impairment or only minor impairment (full support); (2) moderate impairment (partial support); and (3) severe impairment (nonsupport). These impairment levels correspond to excellent, good, fair, and poor biological integrity, respectively.

Quality Assurance

Several steps were taken to assure that the study results are accurate and reproducible. Upon receipt of the samples, station and sample attribute data were recorded in the Montana Diatom Database and the samples were assigned a unique number, e.g., 3045-01. The first part of this number (2951) designates the sampling site (Beaver Creek near mouth) and the second part (01) designates the number of periphyton samples that that have been collected at this site for which data have been entered into the Montana Diatom Database.

Sample observations and analyses of soft (non-diatom) algae were recorded in a lab notebook along with information on the sample label. A portion of the raw sample was used to make duplicate diatom slides. The slide used for the diatom proportional count will be deposited in the Montana Diatom Collection at the University of Montana Herbarium in Missoula. The duplicate slide will be retained by *Hannaea* in Helena. Diatom proportional counts have been entered into the Montana Diatom Database.

Results and Discussion

Results are presented in Tables 3, 4 and 5, which are located near the end of this report following the references section. Appendix A consists of a series of diatom reports, one for each sample. Each diatom report contains an alphabetical list of diatom species and their percent abundances, and values for 65 different diatom metrics and ecological attributes.

Sample Notes

Beaver Creek. All three samples from Beaver Creek were putrid, but the algae were still intact and identifiable. The samples from the upper and lower sites were extremely silty and the sample from the middle site was very silty. The sample from the middle site consisted mostly of detritus. *Cladophora* was present but senescent at all three sites.

Casino Creek. Macroscopically, the specimen in this sample appeared as a large bright green mass of filamentous algae. Microscopically, the mass proved to be a monoculture of *Cladophora*, probably *Cladophora glomerata*, a common filamentous green alga (Division Chlorophyta) that often becomes a nuisance in nutrient-rich lakes and streams in temperate regions around the world. The bright green color indicates fresh new growth; epiphytic diatoms had not yet had a chance to colonize the filaments.

Cottonwood Creek. Macroscopically, the specimen in this sample appeared as a pale yellow to beige flocculent mass about 1 centimeter thick and several centimeters wide and long. Microscopically, the mass proved to be a colony of diatom (Division Bacillariophyta) cells embedded in amorphous mucilage. The cells were mainly a *Cymbella* species and a *Synedra* species. These taxa are known to extrude polysaccharide mucilage through pores in their cell walls, thus forming stalks, tubes, and amorphous masses in which the diatoms live. Certain species of *Cymbella* and *Synedra* are associated with eutrophication.

Non-Diatom Algae (Beaver Creek)

In addition to diatoms, which ranked first in biovolume at all three sites, periphyton samples from Beaver Creek contained cyanobacteria, red algae, green algae, and yellow-green algae (Table 3). At the upper site, the pollution-tolerant cyanobacterium *Oscillatoria* was frequent and ranked 2nd in biovolume. The filamentous green alga *Cladophora* was common and ranked 3rd, while the filamentous red alga *Audouinella* was occasional and ranked 4th.

At the middle site, *Cladophora* and *Oscillatoria* were common and frequent and ranked 2nd and 3rd, respectively, followed in abundance by *Vaucheria* and two genera of green algae: *Oedogonium* and *Closterium*. The chrysophyte *Vaucheria* prefers steady flows of cool water and is often found in springs and spring brooks. At the lower site, *Cladophora* was abundant and ranked 2nd in biovolume, followed by an occasional cell of *Closterium*, which ranked 3rd. *Cladophora* is frequently cited as an aquatic nuisance in the United States (Wehr and Sheath 2003). Large standing crops of *Cladophora* are often a sign of elevated concentrations of inorganic nutrients, particularly phosphorus.

Diatoms (Table 4)

Six of the major diatom species in Beaver Creek are sensitive to organic pollution and these were abundant at all three sites (Table 4). Six of the major species are somewhat tolerant of organic pollution and these were present at all three sites. *Nitzschia linearis*, a somewhat tolerant species that is also highly motile and tolerant of sedimentation, was abundant only at the upper site. Only one of the major diatom species—*Nitzschia palea*—is most tolerant of organic pollution (pollution tolerance class 1). *Nitzschia palea* was most abundant at the upstream site and declined in relative abundance at the middle and lower sites (Table 4).

Diatom metrics suggest the upper site was moderately impaired by sedimentation and provided less than full support for aquatic life uses. This was due to a much larger than normal percentage of motile diatoms, which approached the threshold for severe impairment in a mountain stream. The pollution index was also depressed at the upper site, suggesting minor impairment from organic loading (Table 4). *Nitzschia palea*, a nitrogen heterotroph, was the second most abundant diatom species here. Otherwise, diatom species richness and diversity at this site were acceptable, and no abnormal diatom cells were recorded.

The middle site had the best biological integrity of the three sites on Beaver Creek. Diatom metrics indicate only minor impairment from sedimentation and minor disturbance due to the slightly elevated percentage of *Achnanthidium minutissimum*. The second most abundant species here was *Gomphonema minutum*, an eutraphentic species that suggests elevated levels of inorganic nutrients (Van Dam et al. 1994). Two abnormal diatom cells were recorded at this site, which is normal background. This site shared about half of its diatom assemblage with the upper site, indicating only minor changes in environmental conditions.

A large percentage of motile diatoms suggests the lower site was moderately impaired by sedimentation. *G. minutum* was the dominant diatom species here, indicating eutrophication. The pollution index approached but did not drop below the threshold for minor impairment. No abnormal diatom cells were observed and species richness and diversity were acceptable but on the low side. This site shared about half of its diatom assemblage with the middle site.

Several ecological attributes were selected from the diatom reports in the appendix and modal categories of these attributes were extracted to characterize water quality tendencies at the three sites (Table 5). The majority of diatoms at all three sites were freshwater nitrogen autotrophs that tolerate only a small amount of BOD loading. Nitrogen autotrophs require inorganic nitrogen (nitrates and ammonia) as nutrients.

Most of the diatoms at the middle site were non motile, suggesting that this site had the least amount of sedimentation. On the other hand, the largest motility category at the upper site was "highly motile" and the largest category at the lower site was "moderately motile". The sedimentation index indicated moderate impairment at both of these sites.

The largest trophic state category at the upper site was "variable". Diatoms in this category tolerate a wide range of concentrations of inorganic nutrients. At the middle and lower sites, most diatoms were in the "eutraphentic" category, which suggests that concentrations of inorganic nutrients (carbon, nitrogen, and phosphorus) were elevated at these sites.

Most diatoms at the middle site belong to the "circumneutral" pH category. These diatoms prefer pH levels around 7. At the upper and lower sites, the largest pH category was "alkaliphilous", which suggests higher pH values and perhaps more photosynthesis (primary production) at these sites. The modal category for dissolved oxygen demand was "continuously high" at the upper and middle sites. The largest category for dissolved oxygen demand at the lower site was "not classified".

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Table 1. Location of MDEQ periphyton sampling stations on Beaver, Casino, and Cottonwood Creeks.

Station	MDEQ Station Code	*Hannaea* Sample Number	Latitude	Longitude	Sample Date
Beaver Creek at Regli (upper)	M22BEVRC01	3044-01	46 56 11	109 31 09	7/23/2003
Beaver Creek above county road (middle)	M22BEVRC02	3043-01	46 58 54	109 33 27	7/23/2003
Beaver Creek near mouth (lower)	M22BEVRC04	3045-01	47 04 46	109 35 56	7/24/2003
Casino Creek[1]	M22CSNOC04	[2]	46 59 31	109 27 09	7/23/2003
Cottonwood Creek below Glengarry[1]	M22CTWDC05	3090-01	47 02 24	109 32 41	7/23/2003

[1]These samples were collected only for the identification of conspicuous macroalgae and were not collected following MDEQ standard operating procedures.

[2]A diatom slide was not made from this sample, hence the sample was not assigned a number in the Montana Diatom Database.

Table 2. Diatom association metrics used by the State of Montana to evaluate biological integrity in **mountain** streams: references, range of values, expected response to increasing impairment or natural stress, and criteria for rating levels of biological integrity. The lowest rating for any one metric is the rating for that site.

Biological Integrity/ Impairment or Stress/ Use Support	No. of Species Counted[1]	Diversity Index[2] (Shannon)	Pollution Index[3]	Siltation Index[4]	Disturbance Index[5]	% Dominant Species[6]	% Abnormal Cells[7]
Excellent/None Full Support	>29	>2.99	>2.50	<20.0	<25.0	<25.0	0
Good/Minor Full Support	20-29	2.00-2.99	2.01-2.50	20.0-39.9	25.0-49.9	25.0-49.9	>0.0, <3.0
Fair/Moderate Partial Support	19-10	1.00-1.99	1.50-2.00	40.0-59.9	50.0-74.9	50.0-74.9	3.0-9.9
Poor/Severe Nonsupport	<10	<1.00	<1.50	>59.9	>74.9	>74.9	>9.9
References	Bahls 1979 Bahls 1993	Bahls 1979	Bahls 1993	Bahls 1993	Barbour et al. 1999	Barbour et al. 1999	McFarland et al. 1997
Range of Values	0-100+	0.00-5.00+	1.00-3.00	0.0-90.0+	0.0-100.0	~5.0-100.0	0.0-30.0+
Expected Response	Decrease[8]	Decrease[8]	Decrease	Increase	Increase	Increase	Increase

[1]Based on a proportional count of 400 cells (800 valves)
[2]Base 2 [bits] (Weber 1973)
[3]Composite numeric expression of the pollution tolerances assigned by Lange-Bertalot (1979) to the common diatom species
[4]Sum of the percent abundances of all species in the genera *Navicula, Nitzschia* and *Surirella*
[5]Percent abundance of Achnanthidium minutissimum (synonym: *Achnanthes minutissima*)
[6]Percent abundance of the species with the largest number of cells in the proportional count
[7]Cells with an irregular outline or with abnormal ornamentation, or both
[8]Species richness and diversity may increase somewhat in mountain streams in response to slight to moderate increases in nutrients or sediment

Table 3. Relative abundance of cells and ordinal rank by biovolume of diatoms (Division Bacillariophyta) and genera of non-diatom algae in periphyton samples collected from Beaver Creek near Lewistown in 2003: d = dominant; a = abundant; f = frequent; c = common; o = occasional; r = rare.

Taxa	M22BEVRC01	M22BEVRC02	M22BEVRC04
Cyanophyta (cyanobacteria)			
Oscillatoria	frequent/2nd	frequent/3rd	
Rhodophyta (red algae)			
Audouinella	occasional/4th		
Chlorophyta (green algae)			
Cladophora	common/3rd	common/2nd	abundant/2nd
Closterium		rare/6th	occasional/3rd
Oedogonium		occasional/5th	
Chrysophyta (yellow-green algae)			
Vaucheria		occasional/4th	
Bacillariophyta (diatoms)	abundant/1st	abundant/1st	dominant/1st
Number of Non-Diatom Genera	3	5	2

Table 4. Percent abundance of major diatom species[1] and values of selected diatom association metrics for periphyton samples collected from Beaver Creek near Lewistown in 2003. <u>Underlined values</u> indicate minor stress; **bold values** indicate moderate stress; **<u>underlined and bold</u>** values indicate severe stress; all other values indicate no stress and full support of aquatic life uses when compared to criteria for mountain streams in Table 2. Stress may be natural or anthropogenic (see text).

Species/Metric	PTC[2]	M22BEVRC01	M22BEVRC02	M22BEVRC04
Achnanthidium minutissimum	3	24.82	28.25	0.96
Cocconeis pediculus	3		1.56	3.48
Cymbella excisa	3	3.37	6.25	4.2
Encyonema minutum	2	4.82	1.92	0.96
Gomphonema minutum	3	0.24	27.64	31.06
Navicula capitatoradiata	2	2.53	3.85	26.38
Navicula cryptotenella	2	7.47	0.72	1.68
Navicula tripunctata	3	2.17	1.44	7.67
Navicula trivialis	2	4.10	0.36	0.12
Navicula viridula	2	1.69	0.84	3.96
Nitzschia dissipata	3	9.52	2.16	2.28
Nitzschia linearis	2	6.27	0.48	0.24
Nitzschia palea	1	10.24	4.57	2.88
Number of Species Counted		42	43	37
Shannon Species Diversity		4.11	3.59	3.36
Pollution Index		<u>2.36</u>	2.68	2.54
Siltation Index		**58.80**	<u>23.20</u>	**50.36**
Disturbance Index		24.82	<u>28.25</u>	0.96
Percent Dominant Species		24.82	<u>28.25</u>	<u>31.06</u>
Percent Abnormal Cells		0.00	<u>0.24</u>	0.00
Similarity Index[3]			51.41	52.85

[1]A major diatom species accounts for 3.0% or more of the cells at one or more stations in a sample set.

[2](Organic) Pollution Tolerance Class (Lange-Bertalot 1979): 1 = most tolerant; 2 = tolerant; 3 = sensitive.

[3]Percent Community Similarity (Whittaker 1952) when compared to the diatom assemblage at the next upstream station on Beaver Creek.

Table 5. Modal categories for selected ecological attributes of diatom species in Beaver Creek near Lewistown in 2003. Values that represent inferior water quality when compared to the best site in the sample set are given in **bold** type.

Ecological Attribute	M22BEVRC01	M22BEVRC02	M22BEVRC04
Motility[1]	**Highly Motile**	Not Motile	**Moderately Motile**
pH[2]	**Alkaliphilous**	Circumneutral	**Alkaliphilous**
Salinity[2]	Fresh	Fresh	Fresh
Nitrogen Uptake[2]	Autotrophs (high organics)	Autotrophs (high organics)	Autotrophs (high organics)
Oxygen Demand[2]	Continuously High	Continuously High	Not Classified
Saprobity[2]	beta-Mesosaprobous	beta-Mesosaprobous	beta-Mesosaprobous
Trophic State[2]	Variable	**Eutraphentic**	**Eutraphentic**

[1]Dr. R. Jan Stevenson, Michigan State University, digital communication.
[2]Van Dam et al. 1994